



SEQUENCE LISTING

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<120> Expression Vector For Hirudin And Transformed Cells And
Transgenic Animals Containing Said Vector

<130> MBHB-03-1065

<140> 10/727,145

<141> 2003-12-02

<150> 10/053,641

<151> 2002-01-18

<160> 21

<170> PatentIn version 3.3

<210> 1

<211> 104

<212> DNA

<213> Artificial Sequence

<220>

<223> Single-stranded DNA fragments Hi-AF designed from the hirudin
gene.

<400> 1

gaccccttat ggttggtttac actgactgca ctgaatccgg tcagaacctg tgcctgtgcg 60

aaggtcttaa cgtttgcggc cagggcaaca aatgcacccg gggc 104

<210> 2

<211> 107

<212> DNA

<213> Artificial Sequence

<220>

<223> Single-stranded DNA fragments Hi-AR designed from the hirudin
gene.

<400> 2

ctctagagcc caggatgcat ttgttgccct ggccgcaaac gtttagagcct tcgcacaggc 60

acaggttctg accggattca gtgcagtcag tgtaaacac cataaag 107

<210> 3
 <211> 111
 <212> DNA
 <213> Artificial Sequence

 <220>
 <223> Single-stranded DNA fragments Hi-BF designed from the hirudin gene.

 <400> 3
 tctagaggcg aaaaaaatca atgcgttact ggcgaaggta ctccgaaacc gcagtctcac 60
 aacgacggcg actttgaaga aatcccggaa gaatacctgc aataataggg c 111

 <210> 4
 <211> 108
 <212> DNA
 <213> Artificial Sequence

 <220>
 <223> Single-stranded DNA fragments Hi-BR designed from the hirudin gene.

 <400> 4
 ggccgccccta ttattgcagg tattcttccg ggatttcttc aaagtcgccg tcgttgtgag 60
 actgcggttt cggagtacct tcgccagtaa cgcattgatt tttttcgc 108

 <210> 5
 <211> 33
 <212> DNA
 <213> Artificial Sequence

 <220>
 <223> Primer Hi-PCR-AF designed from the hirudin gene.

 <400> 5
 tcgggatcct ttatggttgt ttacactgac tgc 33

 <210> 6
 <211> 31
 <212> DNA
 <213> Artificial Sequence

 <220>
 <223> Primer Hi-PCR-AR designed from the hirudin gene.

 <400> 6
 gcctctagag cccaggatgc atttgttgcc c 31

 <210> 7
 <211> 38
 <212> DNA

<213> Artificial Sequence

<220>

<223> Primer Hi-PCR-BF designed from the hirudin gene.

<400> 7

ggctctagag gcgaaaaaaa tcaatgcgtt actggcga

38

<210> 8

<211> 32

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer Hi-PCR-BR designed from the hirudin gene.

<400> 8

catgcggccg ccctattatt gcaggtattc tt

32

<210> 9

<211> 45

<212> DNA

<213> Capra hircus

<220>

<221> misc_feature

<223> Signal sequence from a goat beta-casein.

<400> 9

atgaaggtcc tcatccttgc ctgtctggtg gctctggcca ttgca

45

<210> 10

<211> 15

<212> PRT

<213> Capra hircus

<220>

<221> MISC_FEATURE

<223> Signal sequence from a goat beta-casein.

<400> 10

Met Lys Val Leu Ile Leu Ala Cys Leu Val Ala Leu Ala Ile Ala

1

5

10

15

<210> 11

<211> 34

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer Hir1st5' for the adding the signal sequence from a goat

beta-casein to the 5'-terminal of the hirudin gene.

<400> 11
tggctctggc cattgcagtt gtttacaccg actg 34

<210> 12
<211> 34
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer Hir2nd5' for the adding the signal sequence from a goat
beta-casein to the 5'-terminal of the hirudin gene.

<400> 12
tcataccttgc ctgtctggtg gctctggcca ttgc 34

<210> 13
<211> 34
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer Hir3rd5' for adding the signal sequence from a goat
beta-casein to the 5'-terminal of the hirudin gene.

<400> 13
tcgctcgaga tgaaggtcct catccttgcc tgtc 34

<210> 14
<211> 31
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer Hir3'XhoI for adding the signal sequence from a goat
beta-casein to the 5'-terminal of the hirudin gene.

<400> 14
tcgctcgagt tattgcaggt attcttccgg g 31

<210> 15
<211> 243
<212> DNA
<213> *Hirudo medicinalis*

<220>
<221> misc_feature
<223> Sequence of hirudin gene from Genbank accession number of
M12693.

<400> 15

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atgaagggtcc tcatccttgc ctgtctggtg gctctggcca ttgcagttgt ttacaccgac      60
tgactgaat ccggtcagaa cctgtgcctg tgcaaggct ctaacgtttg tggccagggc      120
aacaaatgca tcctgggctc tgacggcgaa aaaaatcaat gcgttactgg cgaaggtact      180
ccgaaaccgc agtctcacia cgacggcgac tttgaagaaa tcccgaaga atacctgcaa      240
taa                                                                    243

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<210> 16
<211> 215
<212> DNA
<213> Hirudo medicinalis

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<220>
<221> misc_feature
<223> Nucleotide sequence of the full-length DNA fragment of the
       complete coding sequence of hirudin.

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<400> 16
gatcctttat ggttggttac actgactgca ctgaatccgg tcagaacctg tgctgtgcg      60
aaggctctaa cgtttgccgc cagggcaaca aatgcctcct gggctctaga ggcgaaaaaa      120
atcaatgcgt tactggcgaa ggtactccga aaccgcagtc tcacaacgac ggcgactttg      180
aagaaatccc ggaagaatac ctgcaataat agggc                                215

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<210> 17
<211> 67
<212> PRT
<213> Hirudo medicinalis

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<220>
<221> MISC_FEATURE
<223> Amino acid sequence of the full-length DNA fragment of the
       complete coding sequence of hirudin.

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<400> 17

Met Val Val Tyr Thr Asp Cys Thr Glu Ser Gly Gln Asn Leu Cys Leu
1          5          10          15

Cys Glu Gly Ser Asn Asn Val Cys Gly Gln Gly Asn Lys Cys Ile Leu
          20          25          30

Gly Ser Arg Gly Glu Lys Asn Gln Cys Val Thr Gly Glu Gly Thr Pro
          35          40          45

Lys Pro Gln Ser His Asn Asp Gly Asp Phe Glu Glu Ile Pro Glu Glu

```

Tyr Leu Gln

65

<210> 18
 <211> 24
 <212> DNA
 <213> Artificial Sequence

<220>

<223> Primer palphaLA-forward for transgene screened by PCR.

<400> 18
 gcttcctaga accaacacta ccag

24

<210> 19
 <211> 21
 <212> DNA
 <213> Artificial Sequence

<220>

<223> Primer palphaLA-Reverse for transgene screened by PCR.

<400> 19
 gtcgccgtcg ttgtgagact g

21

<210> 20
 <211> 27
 <212> DNA
 <213> Artificial Sequence

<220>

<223> Primer pBC1-Forward for transgene screened by PCR.

<400> 20
 gattgacaag taatacgctg tttcctc

27

<210> 21
 <211> 26
 <212> DNA
 <213> Artificial Sequence

<220>

<223> Primer pBC1-Reverse for transgene screened by PCR.

<400> 21
 catcagaagt taaacagcac agttag

26